

**[ The effect of ethanolic leaves extract of *Solenostemma argel* on blood electrolytes and biochemical constituents of albino rats ]**

**Abstract**

The ethanolic leaves extract of *S. argel* was studied for its effect on the blood electrolytes and biochemical constituents of albino rats. The animals (n= 12, average BW= 240 g) were divided into two groups of six each: one to act as the experimental group (EG) and the other as the control group (CG). The result Revealed that K, Ca and P for electrolytes and creatinine, urea, aspartate aminotransferase (AST) and alkaline phosphatase (ALP) for biochemical constituents were found significantly higher ( $P < 0.05$ ) in experimental group when compared with the control group. In the meantime, total protein, albumin, total bilirubin, and alanine aminotransferase (ALT) for biochemical constituents, and Na and Fe for electrolytes did not differ significantly from those of the control group ( $P > 0.05$ ). The findings of this study suggested that the dose 600 mg of extract /Kg BW was more likely to have adverse effect on liver and kidneys of albino rats.



## The effect of ethanolic leaves extract of *Solenostemma argel* on blood electrolytes and biochemical constituents of albino rats

Osman H M<sup>1</sup>, Shayoub M H<sup>2</sup>, Babiker E M<sup>3</sup>, and Mounzer M. Elhag<sup>4</sup>

<sup>1</sup> Department of Biochemistry, Faculty of Pharmacy, National Ribat University,

<sup>2</sup> Department of Pharmaceutics, Faculty of Pharmacy, University of Khartoum,

<sup>3</sup> Department of Zoology, Faculty of Science, University of Khartoum,

<sup>4</sup> Department of Biochemistry, Faculty of Medicine, University of Gadarf.

**Corresponding author:** Dr. Hisham Mohamed Osman, Faculty of Pharmacy, National Ribat University, P.O. Box 55. Mobile No: 912966220. E. mail: [hisham121ribat@yahoo.com](mailto:hisham121ribat@yahoo.com)

### Abstract

The ethanolic leaves extract of *S. argel* was studied for its effect on the blood electrolytes and biochemical constituents of albino rats. The animals (n= 12, average BW= 240 g) were divided into two groups of six each: one to act as the experimental group (EG) and the other as the control group (CG). The result Revealed that K, Ca and P for electrolytes and creatinine, urea, aspartate aminotransferase (AST) and alkaline phosphatase (ALP) for biochemical constituents were found significantly higher ( $P < 0.05$ ) in experimental group when compared with the control group. In the meantime, total protein, albumin, total bilirubin, and alanine aminotransferase (ALT) for biochemical constituents, and Na and Fe for electrolytes did not differ significantly from those of the control group ( $P > 0.05$ ). The findings of this study suggested that the dose 600 mg of extract /Kg BW was more likely to have adverse effect on liver and kidneys of albino rats.

**Key words** = *Solenostemma argel*, medicinal plant, electrolytes, glomerular filtration rate, AST, ALP, ALT, liver function, kidneys function.

### Introduction

The plant *Solenostemma argel* belongs to the *Asclepiadaceae* family. It is an erect shrub reaching a height of 60 to 100cm, with many velvety, pubescent branches from the base. In Sudan, it is indigenous in the northern region between Barbar and Abu-Hamad (Tagelsir, 2011). It is locally known as 'Hargal'. It is widely used in Sudanese traditional folkloric medicine as antispasmodic (Innocenti, *et al.*, 2010), anti-inflammatory and anti-rheumatic agent (Shyoub, *et al.*, 2013). Smoke inhalation and infusion of the whole of this plant is used in treatment of hypercholesterolemia, diabetes mellitus, cold, cough, jaundice and measles. Also, it is described for treating gastrointestinal cramp, urinary tract infection and the menstrual disturbance as well as being used as anti-syphilitic when used for prolonged period (ElKamali and Khalid 1996). The

plant also possesses insecticidal effect and hence was used to combat insect pests (Awad *et al.*, 2012). In this context, it was used against mosquito species, the causative agent of malaria in Sudan (Feiha *et al.*, 2009; Stngeland. *et al.*, 2011; Sameh and Abdelhalim, 2011). Moreover, it was reported to have antimicrobial properties, as well as antibacterial and antioxidant activity (Shafeek and Michael, 2012; Mahalel, 2012).

On other hand, chemical investigations, chromatographic screening and phytochemical as well as tissue culture studies of *S. argel* leaves, stems and flowers revealed the presence of numerous biochemical ingredients and crystalline compounds and most of them were detected when extracted in alcohol such as: Pyrgene glycosides (Hassan *et al.*, 2001; Plaza. *et al.*, 2005), flavonoids, kaempferol (Shafeek and Michael 2012), quercetin, rutin, flavanones, and alkaloids

(Tigani and Ahmed 2009). The leaves of *S. argel* were also characterized by having a high percentage of carbohydrates (64.8), slightly low percentage of protein (15), low percentage of crude fiber (6.5), crude oil (1.6), about 7.7% as ash and 4.4% as moisture. This in addition to lower percentage of minerals namely: potassium (0.54%), calcium (0.06%), magnesium (0.03%), sodium (0.01%), copper (0.0001%), ferrous (0.002%), manganese (0.002%) and lead (0.001%). Furthermore, protein fractionation of leaves revealed albumin percentage of 16.7, non-nitrogenous protein (15.3%), prolamine (11.7%), globulins (8.7 %) and glutulin (6.2%). Together with all these, phytic acid of 3.2 g/100g and tannin content of about 0.4% were reported (Murwan *et al.*, 2010).

The aim of this study is to find out the effect of ethanolic leaves extract of *S. argel* on serum biochemical constituents and electrolytes in albino rats and to discuss their values in relation to kidneys and liver functions.

## Materials & Methods

### Experimental Animals

Albino rats (n= 12, average BW= 240 g) were used in this study and were divided into two groups of six animals; one to act as a control group and denoted (CG) and the other as experimental group and denoted (EG). The members of the control group were provided with normal diet concentrate (dried meat, milk powder, oil and flour in some water) without *S. argel* while those of the experimental group were provided, in addition to the concentrate, with the dose 600 mg/kg of body weight, daily for 30 days.

### Preparation of *S. argel* leaves extract

The leaves of *S. argel* were bought from local market and placed in ethanol (80%) for three days in Soxhlet apparatus. Then the *S. argel* extract was dried in Rotary Evaporator Apparatus, weighed and dissolved in distilled water to prepared the concentration of 600 mg extract /kg of body weight (BW) and, the doses were administrated orally by Gavage for the rats of the experimental group (EG), daily for 30 days.

### Biochemical measurements

For biochemical constituents, about 4 ml of blood sample was collected by hematocrit capillary tube from retro-orbital from each rat. Then the sample was placed in plane tube, left to

coagulate, centrifuged at 1000 rpm and the supernatant (serum) was used for the measurement of electrolytes namely; Na, K, Ca, P and Fe. For Na and K, an aliquot for each element was placed in a cuvette and measured using flame photometer apparatus, and the same was done for measuring Ca, P and Fe using Spectrophotometer apparatus.

On the other hand, total proteins, urea, creatinine, total bilirubin, albumin, ALT, ALP and AST were determined using routinely used kits (Randox Laboratories Ltd, UK).

### Statistical analysis:

Mean values of blood constituents were analyzed by student T. test (Mean  $\pm$  SD), using computer package program (PASW statistics 18).

## Results

The results of serum biochemical constituents and electrolytes are shown in Table 1 and Table 2 respectively. The results indicated that creatinine, urea, AST and ALP had increased significantly ( $P < 0.05$ ) for biochemical constituents and K, Ca, and P had increased significantly ( $P < 0.05$ ) for the electrolytes.

## Discussion

Many studies confirmed that the *S. argel* had remedial effect against numerous diseases and health problems such as diabetes mellitus (Trojan, *et al.*, 2012) and cancer (Amr, *et al.*, 2009; Hanafi and Mansour, 2010). Regarding the latter, pregnane glycosides isolated from this plant were reported to reduce cell proliferation in a dose dependent manner (Plaza. *et al.*, 2005). Nevertheless, in this study the results revealed that the experimental dose of *S. argel* induced high levels of creatinine, urea, alkaline phosphatase (ALP) and aspartate aminotransferase (AST) when compared with those of the control group. It is evident that the levels of these parameters taken together would suggest that *S. argel* had incurred hepatorenal toxicity in the experimental animals. However, similar to this effect was induced in albino rats by alkaloids of *Senna obtusifolia* and *S. alata* (Yagi *et al.*, 1998). Similarly, the presence of these alkaloids in *S. argel* as previously reported (Tigani and Ahmed 2009), would strengthen even more our claim that exceeding dose of *S. argel* had caused the toxic effect on the liver and kidneys of albino rats of this study.

Moreover, concurrent increase in the level of K, Ca and P could furtherly confirm occurrence of toxic effect on the kidneys. In this context, it was reported that hyperkalaemia (increase potassium level) might be caused by many reasons among which are renal damage and cells damage (Anja and Markus, 2011; Eleftheriadis *et al.*, 2012). In addition to reduced renal excretion, excessive intake or leakage of potassium from the intracellular space and acute and chronic renal failure (Anja and Markus, 2011); the potassium released from damaged cells. Furthermore, hypercalcemia revealed in this study was another indicator for the damage inflicted on the renal function of experimental animals. It is obvious that

hypercalcemia develops when the input of calcium to the circulation exceeds its removal by urinary tract (Keiko *et al.*, 2013). In this context, it is also important to be noted that hyperkalemia is itself deleterious to kidneys function, and the reduced glomerular filtration rate is often an important component of any hypercalcemia (Goodman, 2005). Similarly, higher level of Phosphorus detected in this study could support the claim of impairment of the kidneys function by this plant particularly when administered at 600 mg/kg. The occurrence of hyperphosphatemia probably was due to inability of the kidneys to excrete dietary phosphorus load, and it was reported to occur in the kidneys with chronic disease (Tskashi *et al.*, 2012).

Table (1): Mean values of biochemical constituents in both; the control (CG) and experimental group (EG), which received 600 mg/kg of body weight daily for 30 days.

Parameters	CG	EG	Normal range <sup>¶</sup>
Creatinine(mg/dl)	0.2 ± 0.3	0.4 ± 0.1*	0.1 – 0.4
Alb (g/dl)	3.1 ± 0.2	3.2 ± 0.8	2.2 – 3.8
Total protein (g/dl)	6.8 ± 0.4	6.9 ± 0.8	5.7 – 8
Urea (mg/dl)	21±2.8	32.4±1.9**	10 – 32
Total Bilirubin (mg/dl)	0.2 ± 0.7	0.3 ± 3.1	0 – 0.4
(AST) (IU/l)	8.2 ± 1.6	14.1 ±1.2**	1 – 48
(ALT) (IU/l)	5.1 ± 4.1	6.5 ±5.8	1 – 22
(ALP) (IU/l)	75 ± 10	95 ± 9.1**	46 – 245

Values are means ± SD, No = 6. \* = P≤0.05, \*\* = P≤0.01 versus control. <sup>¶</sup> (David *et al.*, 2002).

Table (2): Mean values of electrolytes in both; the control (CG) and experimental group (EG), which received 600 mg/kg daily for 30 days.

Parameters	CG	EG	Normal range <sup>¶</sup>
Na (mmol/l)	155.2 ± 2.8	156.2 ± 1.5	145 – 155
K (mmol/l)	3.1 ± 0.2	4.5 ± 0.3*	3.5 – 4.8
Fe (μmm/l)	3.8±2.6	3.9±3.3	3.5 – 4.5
Ca (mg/dl)	6.4± 0.2	8.7 ±4.9**	7.7 – 12
P (mg/dl)	5.8±1.1	6.9±3.5**	4 –10.2

Values are means ± SD, No = 6. \* = P≤0.05, \*\* = P≤0.01 versus control. <sup>¶</sup> (David *et al.*, 2002).

On the other hand, retention of high levels of urea and creatinine as well as electrolytes was probably due to marked reduction in the glomerular filtration capacity of the kidneys. In this context, it

was reported that the increase in serum urea and creatinine are always associated with the reduction in glomerular filtration capacity (Guy and Wim,

2008) and, therefore, are used as indicator for nephrotoxicity (Tobias and Philip, 2011).

Likewise, secretions of higher levels of alkaline phosphatase (ALP) and aspartate aminotransferase (AST) are used as indicator to hepatocellular damage inflicted on the liver (Shivaraj, *et al.*, 2009). Therefore, elevation in levels of these enzymes in the blood circulation is thought to occur as a result of a large number of disorders such as a gallstone or tumor blocking the common bile duct, alcoholic liver disease or drug induced hepatitis that blocks the flow of bile in smaller bile channel within the liver (Sanjay, *et al.*, 2004).

Extrapolating the findings of the present study to human use of *S. argel*, it could be of significance to propose for those seeking *S. argel* for treatment, to use the plant with the dose far below 600 mg/Kg and to monitor closely the levels of mentioned indicators during the course of treatment.

## References:

Amr, A. N., Ahmed, M. A., Khalid, M. A., David A. L., Alan, C. and Hany, A. E. Anti-cancer and anti-oxidant activity of some Egyptian medicinal plants, *Journal of Medicinal Plants Research* . 3(10): 799 –808 (2009).

Anja, L. and Markus, J. Pathogenesis, diagnosis and management of hyperkalaemia. *Pediatr Nephrol*. 26(3): 377–384 (2011).

Awad, K. T., Khalid, O. A., Tagelsir, I. M. and Sidahmed, O. Argel (*Solenostemma argel* Del. Hayenne) applications for control of the date palm green scale insect (*Asterolicanium phoenicis* Rao) and yield enhancement. *ARPN Journal of Agricultural and Biological Science*.7:6 (2012).

Eleftheriadis, T., Leivaditis, K., Antoniadi, G. and Liakopoulos, V. Differential diagnosis of hyperkalemia: an update to a complex problem. *Hippokratia*. 16(4): 294 – 302 (2012).

ElKamali, H. H. and Khalid, S. A. The most common herbal remedies in central Sudan. *Fitoterapia*. 4: 301 (1996).

Feiha, M. H., Awad, K. T., Hatim, G. and Omar A. Water extracts of Hargal plant (*Solenostemma argel*, Del Hyne) and Usher (*Calotropis procera* Ail) leaves as natural insecticides against mosquito larvae. *J.Sc. Tech*. 10 (3):67 (2009).

Goodman, W. G. Calcium and phosphorus metabolism in patients who have chronic kidney disease. *Med Clin North Am*. 89: 631– 647 (2005).

Guy, D. and Wim, M. Clinical laboratory evaluation of the syndrome of inappropriate secretion of antidiuretic hormone. *CJASN*. 3(4): 1175-1184 (2008).

Hanafi, N. and Mansour, S. Antitumor efficacy of *Solenostemma Argel* and/or  $\gamma$ -irradiation against ehrlich carcinoma. *Journal of Biological Sciences*. 10(6): 468 – 479 (2010).

Hassan, H., Hame, A., El-Emary, Springue, I., Mitome, H. and Miyaoka, H. Pregene derivatives from *Solenostemma argel* leaves. *Pytochemistry*. 57(4):507 – 511 (2001).

Innocenti, G., Dall'Acqua, S., Minesso, P., Budriesi, R., Micucci, M. and Chiarini, A. Evaluation of muscarinic M3-receptor antagonism of *Solenostemma argel* leaves. *Planta Med*. 76: 634 (2010).

Keiko, A., Jonathan, B., David, C. and Nazanene, H. Hypercalcemia in upper urinary tract urothelial carcinoma: A case report and literature review. *Case Rep Endocrinol*. 2013: 470890 (2013).

Mahalel, U.A. Antibacterial sensitivity for some chemically diverse steroid glycosides in vitro. *J. Agric. Soc. Sci.*, 8: 24–28 (2012).

Murwan, K., Sabah, E., Murwa, A. Chemical composition, minerals, protein fractionation, and anti-nutrition factors in leaf of Hargel plant (*Solenostemma argel*). *Euro. J. Sci. Re*. 43(3): 430 – 434 (2010).

Plaza, A., Perrone, A., Balestieri, M., Felice, F., Balestrieric, C. New unusual pregnane glycosides with anti proliferative activity from *Solenostemma argel*. *Steroids*.70(9):594-603 (2005).

Sameh, F. A. and Abdelhalim, A. M. Survey on medicinal plants and spices used in Beni-Sueif, Upper Egypt. *Ethnobiol Ethnomed*. 7: 18 (2011).

Sanjay, K., Kenneth, B., Johan, P., Lawrence, B. Histologic changes mimicking biliary disease in liver biopsies with venous outflow impairment. *Modern Pathology*. 17(7):874 – 878 (2004).

Shafeek, R. E. and Michael, H. N. Antibacterial and antioxidant activity of two new kaempferol glycosides isolated from *Solenostemma argel* stem extract. *Asian journal of plant sciences*. 11 (3): 143 – 147(2012).

Shivaraj, G., Parkash, B., Vinayak, V., Avinash, A., Math, V. and Shruthi, K. A review on laboratory liver function tests. *Pan Afr Med J.* 3: 17 (2009).

Shayoub, M., Haj, E., Makawy, A., Rasha, R. Mona, A. Adverse reaction of *Solenostemma argel* leaves, extraction and alkaloids tablets administered to patients. *Global J Trad Med Sys.* 2(1): 14-18 (2013).

Stngeland, T., Alele, P. E., Katuura, E. and Lye, K. A. Plants used to treat malaria in Nyakayojo sub-county, western Uganda. *J Ethnopharmacol.* 137(1):154-166 (2011)

Tagelsir, I. M., Asma, M. A., Elfatih, M. Mahdi and Awad K.T. Influence of argel (*Solenostemma argel* Del. Hayne) soil applications on flowering and yield of date palm (*Phoenix dactylifera* L.). *Agric. Biol. J. N. Am.* 2(3): 538-542 (2011).

Tigani, S. and Ahmed, S. *Solenostemma argel* tissue culture for production of secondary metabolites. *JGEB.* 7(1): 20 – 23 ((2009).

Tobias, C. and Philip, W. Biomarkers for drug-induced renal damage and nephrotoxicity — an overview for applied toxicology. *AAPS J.* 13(4): 615 – 631 (2011).

Trojan-Rodrigues, M., AAlves, T. L., Soarer, G. L. and Ritter, M. R. Plants used as anti diabetics in popular medicine in Rio Grande do Sul, southern Brazil. *J Ethnopharmacol.* 139(1):155-163 (2012).

Tskashi, S., Yuri, N., Masaki, O., Koichi, T., Daisuta, K. The management of hyperphosphatemia by lanthanum carbonate in chronic kidney disease patients. *Int J Nephrol Renovasc Dis.* 5: 81–89 (2012).

Yagi, S. M., Tigani, S. E., Adam, S. E. Toxicity of *Senna obtusifolia* fresh and fermented leaves (kawal) *Senna alata* leaves and some products from *Senna alata* on rats. *Phytotherapy Research.* 12: 324 – 33 (1998).